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## Effects of Total Phosphorus (TP) and Microbially Available Phosphorus (MAP) on Bacterial Regrowth in Drinking Water Distribution System

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### Abstract

Changes of total phosphorus (TP) and microbially available phosphorus (MAP) in the drinking water distribution system and its' effects on bacterial regrowth were investigated in a city for environment engineering. The content of TP was 2.93~21.66μg/L and the content of MAP was 0.69~8.01μgPO<sub>4</sub><sup>3-</sup>-P/L in the network. They decreased a little with the extend of hydraulic retention time. They also increased in some sampling point by cooperation of suspended bacteriam, biofilm and granule in the drinking water distribution system. Although an initial conclusion that phosphorus was a factor influencing the growth of bacteria in some sampling points by comparison of AOCpotential, AOC<sub>p</sub> and AOCnative had been deduced. Phosphorus was not the key factor that control bacterial regrowth in whole network because of high content of TP and MAP.

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**Keywords** biostability, criteria, bacterial regrowth, total phosphorus (TP), microbially available phosphorus (MAP), environment engineering; drinking water distribution system

### 1. Introduction

Bacteria multiply in the network will increase the possibility of the outbreak of infectious disease by water and makes sensual water quality index becomes poor. Usually we control the bacterial regrowth through chlorine disinfection. However, increasing chlorine dosage will makes the taste of water becomes poor, at the same time it will produce disinfection by-products (DBPs) which are harm to human health. For a long time, organic carbon has been considered as the most primary nutrients to control heterotrophic bacteria to grow in drinking water. Especially the assimilable organic carbon (AOC) that is prone to microbial degradation is considered as the main index of evaluating biological stability of drinking water. But in recent years, some research indicates that in the northern hemisphere (such as Europe, Russia and North America), natural water (e.g. lakes, rivers, and even groundwater) contains a lot of organic carbon as the forest and peat exist and the microbial growth in drinking water was not restricted by AOC, but the Inorganic element phosphorus.

This paper takes the drinking water distribution system as the research object. Through sampling and analysing the contents and changes of total phosphorus (TP) and microbially available phosphorus (MAP) in the water as well as detection of bacterium quantity, it can study the effects of total phosphorus (TP) and microbially available

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phosphorus (MAP) on bacterial regrowth in the network.

## 2. Materials and methods

### 2.1. Sample sites in J waterworks

Testing samples is taken from J waterworks and its feed mains. J waterworks adopts the traditional process (coagulation, precipitation, filtration, chlorine disinfection). We select 11 sampling points in its drinking water distribution system. The distance between all sampling points and J waterworks and hydraulic retention time see table 1. Sampling bottles is fine mouth bottle, Pretreatment methods include pickling, rinse with pure water, processing in muffle furnace of 500 °C without carbonization. Samples are collected in preconditioned sampling bottles, if water contains surplus chlorine, add right amount Sodium thiosulfate (according to the mole ratio Sodium thiosulfate residual chlorine = 1.2:1) to neutralize the residual chlorine in water. All samples should be returned to laboratory within 7h, and Tested within 24h. Analysis of the project mainly includes: TP, MAP, HPC, AOC, etc.

Table 1 Length between sample sites and the plant, hydraulic retention time

numbers	Hydraulic retention time (h)	distance (km)	diameter (mm)
1#	9.00	8.8	200
2#	8.64	7.3	200
3#	11.00	9.0	200
4#	12.56	10.0	200
5#	16.31	12.9	200
6#	19.34	11.1	200
7#	16.40	11.7	700
8#	19.50	15.4	700
9#	5.51	3.4	600
10#	8.46	4.7	600
11#	11.40	6.0	600

### 2.2. The measuring method of total phosphorus

It is the Molybdenum antimony anti spectrophotometric method that was adopted to measure the total phosphorus in test samples. And potassium persulfate was used for dissolution, 752 type spectrophotometer for detection. To improve the measuring accuracy, we choose the cuvette of 50 mm optical path.

### 2.3. The measuring method of microbially available phosphorus

Finland scholars Lehtola etc advances the measuring method of microbially available phosphorus (MAP) in water. Through conducting the research to it, the author confirms the Yield coefficient that the text will use, and dose some improvement to the measuring method for reducing measuring workload. When measure the MAP in water sample, Take 50mL water samples into a Grinding mouth triangle flask of 50 ml which has been processed without carbonization. add enough carbon and other inorganic nutrition except phosphorus into the being tested samples so that the limit of carbon and other inorganic element to microbial growth can be eliminated. Then phosphorus become the only nutrient elements that limit the microbial growth. Add 50  $\mu\text{L}$  Inorganic nutrients (nitrogen, magnesium, calcium, potassium, etc.) and 50  $\mu\text{L}$   $2\text{g}\cdot\text{L}^{-1}$  Acetic acid sodium solution (With Acetic acid sodium Plan) into the 50 ml water samples to make the concentration of the adding nutrients as follows:  $250\mu\text{g}\cdot\text{L}^{-1}\text{N}$ 、 $53\mu\text{g}\cdot\text{L}^{-1}\text{K}$ 、 $10\mu\text{g}\cdot\text{L}^{-1}\text{Mg}$ 、 $27\mu\text{g}\cdot\text{L}^{-1}\text{Ca}$ 、 $40\mu\text{g}\cdot\text{L}^{-1}\text{Na}$ 、 $157\mu\text{g}\cdot\text{L}^{-1}\text{Cl}$ 、 $2000\mu\text{g}\cdot\text{L}^{-1}$  Acetic acid sodium. In the 70 °C water-bath, pasteurize 30min to damage the plant cell and inactivate the spores bacteria, then cool the water bath to

room temperature, according to the Vaccination concentration of  $10^3$  CFU / mL, inoculat pseudomonas fluorescens P 17, train In 15 °C in dark, and count the p17 in the water sample on the third and forth day of cultivation respectively through plate count method. Take maximum number of bacterial colonies in the third and forth day and use yield coefficient calculate the MAP in sample. The yield coefficient for the MAP used in the test is  $1.10 \times 10^9$  each· $\mu\text{g}^{-1}$ .

#### 2.4. Heterotrophic Plate Counting (HPC)

HPC adopt R2A medium, cultivate 7d in 22 ~ 28 °C. R2A medium is widely used in international water study .Its nutrient composition is relatively comprehensive, available for many widespread bacterial growth in water, and because HPC cultivation need a long period of time, many slow-growing bacteria also can be detected. Therefore, the results of HPC can reflect heterotrophic bacteria quantity in drinking water better.

### 3. Results and discussion

#### 3.1. Effects of total phosphorus on suspending bacterial regrowth in drinking water distribution system

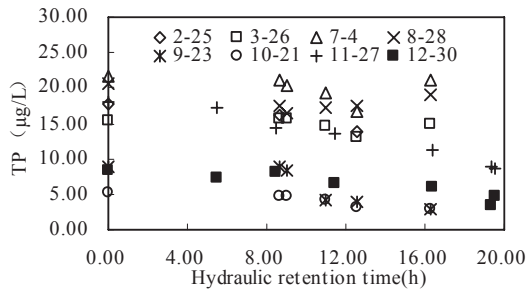
As shown in Table 2 (Figure 1) were Changes of TP with hydraulic retention time in different sampling time (month-day).The content of TP was 2.93~21.66 $\mu\text{g/L}$  in the network, and it was a little higher. the content of TP in the network varied a lot , especially in July and August hydraulic the content of TP was the highest. This was because Source water in J waterworks was reservoir water, and rainfall was more in summer so that surface runoff takes non-point phosphorus pollution and lead to the content of TP increase in Source water. The content of TP in March and November were both higher, also because of rainfall before Sampling. The content of TP was lower in other months. We can see that phosphorus in surface waters mainly comes from non-point pollution of the surface.

From table 2, we can see that the content of TP in the network decreases a little with the extend of hydraulic retention time. This may be due to the two kinds of cause, firstly, The content of TP was consumed constantly by free bacteria and biofilm ;Secondly, TP were adsorbed by particles in the network and biofilm causing the content of TP to reduce. But in individual sampling points TP increase, this might be relevant to the particles which were formed by biofilm desorption.

Table 2 Changes of TP with hydraulic retention time in different sampling time (month-day)

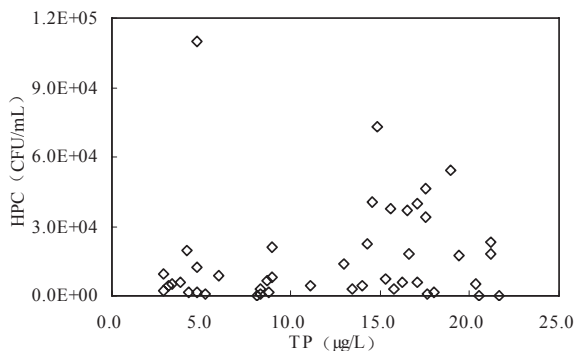
hydraulic retention time (h)	TP ( $\mu\text{g/L}$ )							
	2-25	3-26	7-4	8-28	9-23	10-21	11-27	12-30
0.00	17.66	15.27	21.66	20.49	8.97	5.21	18.01	8.30
5.51	—	—	—	—	—	—	17.10	7.38
8.46	—	—	—	—	—	—	14.29	8.11
8.64	16.23	15.72	21.21	17.5	8.93	4.73	—	—
9.00	16.25	15.55	20.29	16.52	8.33	4.76	—	—
11.00	—	14.55	19.38	17.09	4.30	4.19	—	—
11.40	—	—	—	—	—	—	13.47	6.41
12.56	13.95	12.98	16.64	17.55	3.84	3.19	—	—
16.31	12.80	14.81	21.21	18.92	2.93	2.93	—	—
16.40	—	—	—	—	—	—	11.10	6.01
19.4	—	—	—	—	—	—	8.76	3.38
19.50	—	—	—	—	—	—	8.64	4.76

“—” Note: no sampling



**Fig.1** Changes of TP with hydraulic retention time in different sampling time (month-day)

Figure 2 showed effects of total phosphorus on suspending bacterial regrowth in drinking water distribution system. Overall, the higher the content of total phosphorus in the network was, the more the number of bacteria (HPC) was. Correlation coefficient between TP and HPC was 0.137 ( $n = 45$ ), and the correlation was poor. It showed that correlation was poor through calculating the Correlation coefficient( $r$ ) between the average TP of each different sampling point, maximum TP and the average HPC of each different sampling point, maximum HPC. This explained that the total phosphorus was not directly related to the suspended bacterial growth in the network, namely TP was not the key factor that affect the bacterial regrowth. This may be because TP detected by water sample included not only microbially available phosphorus but also included phosphorus that cannot be available by Microorganisms. The growth of bacteria was restricted by microbially available phosphorus, and irrelevant with other forms of phosphorus.



**Fig.2** Relation between TP content and HPC in the network

### 3.2. Effects of MAP on suspending bacterial regrowth in drinking water distribution system

As shown in table 3 (Figure 3) were Changes of MAP with hydraulic retention time in different sampling time (month-day).the content of MAP was  $0.69 \sim 8.01 \mu\text{gPO}_4^{3-}\text{-P/L}$  in the network, and it was a little higher. The variation rules of MAP was different from The variation rules of TP in different sampling time. The largest concentration of different MAP appeared in November and it was also common higher in December. The Phenomenon showed that the ratio of MAP and TP was variational for different water sample, and when the content of TP was high, the content of MAP was not necessarily high. what's more, we couldn't substitute TP for MAP. It also explained that results of 3.1 experiment why the correlation of TP and HPC was poor.

It can be seen that the MAP decreased gradually with the extend of hydraulic retention time from table 3. Similarly, there also exist the phenomenon of MAP rising in the network. This may be due to the cooperation of free bacteria, biofilm and granule.

Table 3 Changes of MAP with hydraulic retention time in different sampling time (month-day)

hydraulic retention time (h)	MAP ( $\mu\text{gPO}_4^{3-}\text{-P/L}$ )						
	3-26	7-4	8-28	9-23	10-21	11-27	12-30
0.00	4.31	6.31	3.68	7.03	4.11	8.01	5.31
5.51	—	—	—	—	—	7.93	4.13
8.46	—	—	—	—	—	5.50	4.67
8.64	3.36	4.24	2.63	5.24	2.44	—	—
9.00	3.33	3.19	2.69	6.13	1.11	—	—
11.00	3.19	2.70	2.72	2.82	1.19	—	—
11.40	—	—	—	—	—	4.22	3.76
12.56	2.76	2.90	2.69	2.03	1.69	—	—
16.31	4.68	3.10	0.69	1.85	1.83	—	—
16.40	—	—	—	—	—	3.35	3.22
19.34	—	—	—	—	—	3.59	2.82
19.50	—	—	—	—	—	3.10	3.25

“—” Note: no sampling

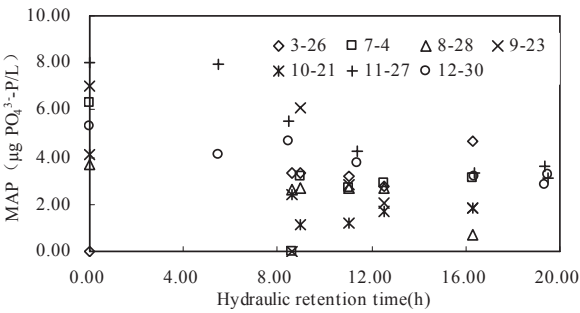


Fig.3 Changes of MAP with hydraulic retention time in different sampling time (month-day)

Figure 4 showed effects of MAP on HPC in drinking water distribution system, HPC has increasing trends with the increasing of MAP.

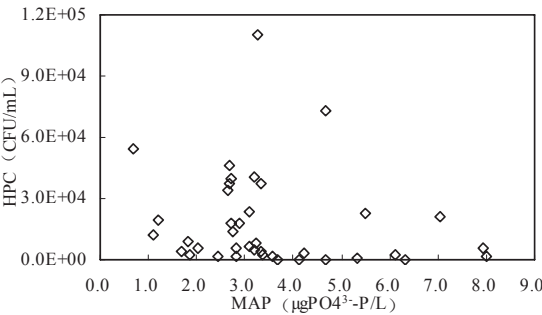


Fig.4 Relation between MAP content and HPC in the network

There was no definite correlation between MAP and HPC through calculating ( $r = -0.114$ ,  $n = 42$ ), it showed that correlation was poor through calculating the Correlation coefficient( $r$ ) between the average MAP of each different sampling point, maximum MAP and the average HPC of each different sampling point, maximum HPC. This explained that MAP was not the key factor that affect the suspending bacterial regrowth.

### 3.3. Discussion

We did some research on the limit of phosphorus to bacteria in some sample points in the earlier time of the study, and found there exist the case that phosphorus limited bacterial growth in some sample points in the network by comparing AOCpotential, AOCp and AOCnative. Through 3.2 analysis, suspended bacterial growth was not limited by phosphorus in the network, so phosphorus is not the main limiting factor. The content of MAP was  $0.69 \sim 8.01 \mu\text{gPO}_4^{3-}\text{-P/L}$  in the network, although it was a little lower, it was enough to Provide phosphorus source that microbial growth needed. The related research put forward when phosphorus was  $1\text{-}3 \mu\text{g P/L}$ , even lower than  $1 \mu\text{g P/L}$  in water, Phosphorus would be the key factor that control bacterial regrowth in whole network. In addition, as the content of TP was  $2.93 \sim 21.66 \mu\text{g/L}$  on average in the network, it was enough to meet the need of suspended bacteria growth. So phosphorus couldn't be the Limiting factor. Overall, there exist the phenomenon that phosphorus limited bacterial growth in some network, but on the whole, the content of TP was still high enough to maintain bacterial growth in the network and phosphorus is the main factor that control bacterial growth.

### 4. Conclusion

The content of TP and MAP decreased a little with the extend of hydraulic retention time. They also increased in some sampling point by cooperation of suspended bacterium, biofilm and granule in the drinking water distribution system. The content of TP was  $2.93 \sim 21.66 \mu\text{g/L}$  and the content of MAP was  $0.69 \sim 8.01 \mu\text{gPO}_4^{3-}\text{-P/L}$  in the network. And phosphorus was not the key factor that control bacterial regrowth in whole network because of high content of TP and MAP.

### References

1. Singer P C. Humic substances as precursors for potentially harmful disinfection by-products. *Water Science and Technology*, 1999, 40(9): 25-30
2. Lehtola M J, Miettinen I T, Hirvonen A, et al. Estimates of microbial quality and concentration of copper in distributed drinking water are highly dependent on sampling strategy. *Int J Hyg Environ Health*, 2007, 210: 725-732
3. Srinivasan S, Harrington W G. Biostability analysis for drinking water distribution systems *Water Research*, 2007, 41: 2127-2138
4. LeChevallier M W, Schulz W, Lee R G. Bacterial nutrients in drinking water. *Appl Environ Microbiol*, 1991(57): 857–862
5. Peter M Huck. Measurement of biodegradable organic matter and bacterial growth potential in drinking water: a review [J]. *J AWWA*, 1990, 82(7): 78-86
6. Lehtola M J, Miettinen I T, Vartiainen T, et al. Changes in content of microbially available phosphorus, assimilable organic carbon and microbial growth potential during drinking water treatment processes. *Water Research*, 2002, 36: 3681-369
7. NEPA. 《Water and wastewater inspection standard》 Editorial committee. *Water and wastewater inspection standard*[M]. Beijing: China architecture & building press, 2000
8. Lehtola M J, Miettinen I T, Vartiainen T, et al. A new sensitive bioassay for determination of microbially available phosphorus in water. *Appl Environ Microbiol*, 1999, 65(5): 2032-2034
9. Jang Dengling, LuWei, Zhang Xiaojian. Research on the measuring method for microbially available phosphorus (MAP) [J]. *water supply and sewage*, 2004, 30(4):27-31
10. Reasoner D J, Geldreich E E. A New Medium for the Enumeration and Subculture of Bacteria from Potable Water [J]. *Appl Environ Microbiol*, 1985, 49(1): 1~7
11. Jang Dengling, Zhang Xiaojian. The relationship of phosphorus and bacterial regrowth in a city.[J]. *Environmental science*, 2004, 25(5):57-60
12. Sathasivan A and Ohgaki S. Application of new bacterial regrowth potential method for water distribution system - a clear evidence of phosphorus limitation [J]. *Water Research*, 1999, 33(1): 137-144